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Increased Levels of the Soluble Adhesion Molecule E-Selectin in Patients With Chronic Myeloproliferative Disorders and Thromboembolic Complications

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Patients with chronic myeloproliferative disorders (CMD) show a high frequency of thrombosis. For this reason we evaluated endothelial cell markers, soluble adhesion molecule E-selectin (sELAM), and thrombomodulin (TM) in 25 patients with CMD. Among them nine presented thromboses in their past history. Data were compared with those obtained in a group of healthy subjects and a group of patients with secondary thrombocytosis.

The mean plasma concentrations of sELAM were elevated in patients with CMD, as compared with healthy subjects (81.27 ± 42.8 ng/ml vs. 41.75 ± 13; $P < 0.02$). Similarly, the mean plasma concentrations of sTM were increased in CMD patients in comparison with the control group (102.0 ± 73 ng/ml vs. 16.7 ± 9.6; $P < 0.01$). More markedly elevated sELAM levels were observed in CMD patients with thrombosis than in patients without thrombosis (113.16 ± 29.5 ng/ml vs. 55.11 ± 19.1 ng/ml; $P < 0.001$), while no significant difference was found between CMD patients without thrombosis and secondary thrombocytosis (50.72 ± 10.8 ng/ml). Plasma thrombomodulin values in CMD patients with thrombosis (131 ± 93.8 ng/ml) were higher than those without thrombosis (65.77 ± 43.9 ng/ml; $P < 0.02$). sTM values were also significantly increased in patients with secondary thrombocytosis ($P < 0.01$).

It is speculated that the plasma, sELAM levels may reflect endothelium activation and that it is possibly useful in predicting the thrombotic risk in myeloproliferative disorders. Am. J. Hematol. 57:109–112, 1998.

Key words: myeloproliferative disorders; endothelial damage; soluble E-selectin; soluble thrombomodulin

INTRODUCTION

Thromboembolic complications are common in patients with chronic myeloproliferative disorders (CMD) [1]. The pathogenesis of these complications is not completely understood, and neither platelet count nor other factors like platelet in vitro function or blood hyperviscosity have been correlated with them [2–4]. It is also interesting to note that patients with secondary thrombocytosis do not have an increase in thromboembolic complications [3], so risk factors other than platelets must contribute in determining thrombotic episodes in subjects with CMD. Loss of the thromboresistant properties of intact endothelium [5,6] could account for the increased incidence of thrombosis, and the evaluation of the degree of vascular endothelial damage could reflect the risk of thrombotic complications in CMD patients. Recently, several vascular endothelial cell markers such as tissue plasminogen activator, von Willebrand factor, and others have been proposed to detect dysfunctional endothelium [7].

In this study, we examined the plasma levels of soluble E-selectin (sELAM) and soluble thrombomodulin (sTM) in CMD patients, and investigated their relationship with thromboembolic complications.

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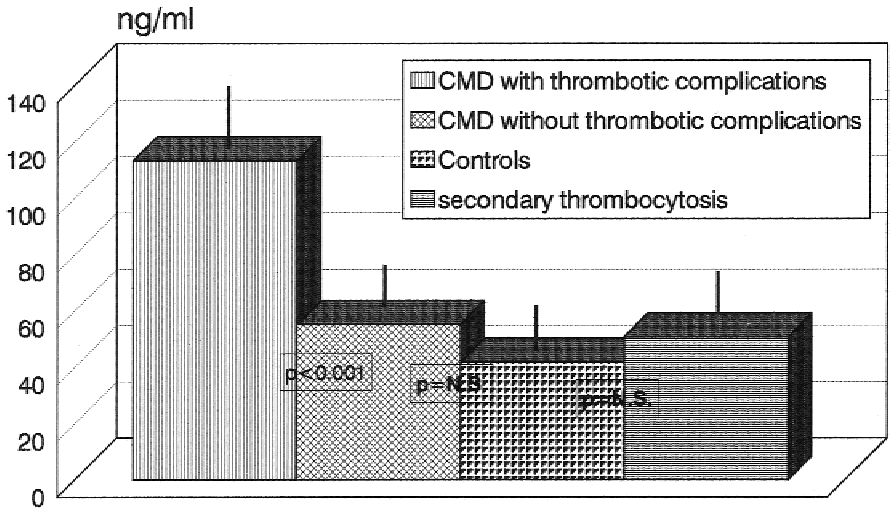

Fig. 1. Plasma sELAM levels of CMD patients with and without thrombotic complications, control subjects, and patients with secondary thrombocytosis.

MATERIALS AND METHODS

The subjects consisted of 25 patients (10 M, 15 F; mean age 54 ± 12 years) with different myeloproliferative disorders (16 essential thrombocythemia, 6 polycythemia vera, 2 chronic myelogenous leukemia, 1 myelofibrosis). The diagnosis was made according to established criteria including bone marrow biopsy. Patients were not on antiplatelet, aspirin, or cytoreductive therapy (alkylating agents or venosection) during the study period and for at least 4 weeks prior. Patients with hypercholesterolemia, diabetes, liver, or kidney disease were excluded. Parallel to the investigation of the CMD patients, 10 healthy control subjects and 5 patients with secondary thrombocytosis to acute cerebrovascular diseases were studied. They were selected to be of the same sex and age of the patients. The CMD patients were also separated into two different groups: with (9 patients: 5 essential thrombocytemia, 2 polycythemia, 1 myelofibrosis, and 1 chronic myelogenous leukemia) or without (16 patients: 11 ET, 4 PV, 1 CML) thromboembolic complications occurring from 1 month prior to the initial diagnosis of CMD to the time of testing. Eight patients were in chronic-phase thrombosis (8 ± 4 months), while 1 patient was in acute-phase thrombosis (3 days after a deep venous thrombosis). Thromboembolic complications included complete stroke, deep venous thrombosis, myocardial infarction, and documented venous arteria occlusions. Angina pectoris and transient ischemic episodes were not included. All patients, with thrombosis or without thrombosis, were in the chronic disease phase.

Venous blood samples were taken after overnight fasting and platelet counts were immediately measured using an electronic counter. Platelet poor plasma was stored at −20°C until assayed. Soluble E selectin was measured with an immunoenzymometric technique (human soluble E selectin immunoassay, R&D System, Abingdon, UK). Plasma thrombomodulin was quantitated with commercial ELISA (STAGO, France).

Data were expressed as mean ± SD. Statistical analysis was performed by using the ANOVA one-way test and Pearson's coefficient of correlation. The significance level was set to $P < 0.05$.

RESULTS

In the CMD patients, the plasma sELAM was significantly elevated compared to the healthy patients (81.27 ± 42.8 ng/ml vs. 41.75 ± 13; $P < 0.02$). The CMD patients with thrombosis showed higher values of sELAM compared to the CMD patients without thrombosis (113.16 ± 29.5 ng/ml vs. 55.11 ± 19.1; $P < 0.001$), the healthy subjects ($P < 0.001$), or those with secondary thrombocytoses (50.72 ± 10.8 ng/ml; $P < 0.001$). sELAM levels showed no significant difference between the CMD patients without thrombosis, the healthy subjects, or those with secondary thrombocytoses, and between those with secondary thrombocytoses and the healthy subjects (Fig. 1).

sTM Levels were higher in the CMD patients than in the healthy subjects (102.0 ± 73.0 ng/ml vs. 16.7 ± 9.6; $P < 0.01$). The CMD patients with thrombosis showed higher values of sTM compared to those without thrombosis (131.0 ± 93.8 ng/ml vs. 65.77 ± 43.9; $P < 0.02$), and the healthy subjects ($P < 0.01$), while no difference was found with the secondary thrombocytoses patients (48.75 ± 18 ng/ml; *P*, NS). sTM levels were significantly increased in the CMD patients without thrombosis in comparison to the healthy subjects ($P < 0.01$), and in the patients with secondary thrombocytoses compared to the healthy subjects ($P < 0.01$) (Fig. 2).

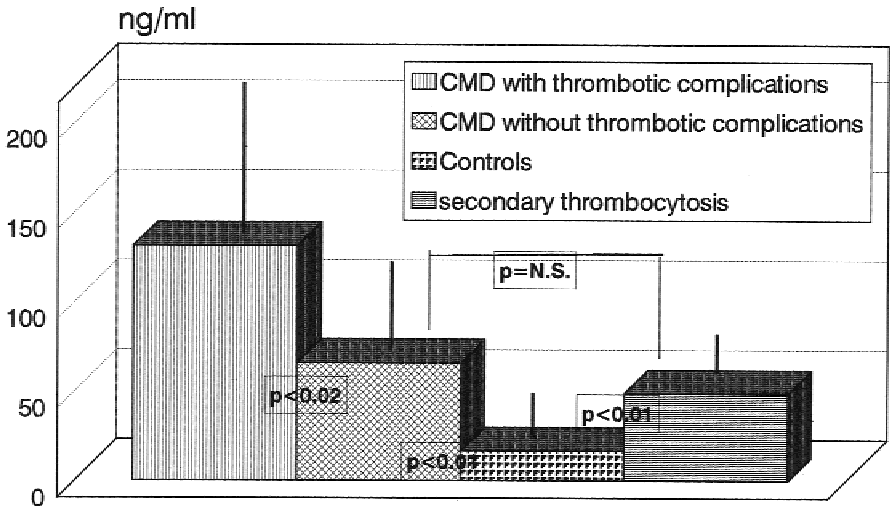

Fig. 2. Plasma thrombomodulin levels of CMD patients with and without thrombotic complications, control subjects, and patients with secondary thrombocytosis.

The plasma sELAM levels were not correlated with sTM in any of the groups studied. Neither sELAM levels nor sTM concentrations were correlated with platelet count.

DISCUSSION

E-selectin is an adhesion molecule that mediates contact between endothelial cells and other cells. Normal resting endothelial cells do not express E-selectin, but a soluble form of this molecule is released from activated endothelial cells. Raised plasma levels have been reported in cancer, hypertension, diabetes, and septic shock [8–10].

Thrombomodulin (TM) is a regulator of activated thrombin, and a soluble form of TM was also used for evaluating the endothelial damage [11]. Increased levels of sTM have been reported in chronic myelogenous leukemia [12], thrombotic thrombocytopenic purpura [13], and DIC [14].

Our data confirm the existence of vascular endothelial damage in patients with CMD that can be disclosed by measuring sELAM and sTM. Although both molecules were higher in the CMD patients compared with the control group, and their plasma levels were higher in the CMD patients with thrombotic lesions than with the patients without thrombotic complications, their behaviour seems quite different. Increased sELAM concentration would appear to be a particularly useful tool to distinguish CMD patients with thrombotic events as its level is raised only in such patients, unlike what occurs with sTM whose values are also higher in CMD patients without thrombotic lesions and in subjects with secondary thrombocytoses. In fact sE-selectin dosage is particularly interesting because this molecule is only found on activated endothelium [8], in contrast to TM, which has a wider distribution. Demonstration of sELAM in the blood could therefore be taken as conclusive evidence of endothelium activation. In contrast, sTM levels in the blood are probably not endothelium specific because TM is also found in other cells such as platelets, in particular, and also in placental syncytiotrophoblasts [15,16].

Our data could, therefore, be useful to improve our understanding of the pathogenic causes behind thrombotic events in patients with chronic myeloproliferative disorders. It is possible that the state of endothelial activation or damage, probably present in these patients, could be more important in determining thromboembolism than other situations such as the number of platelets or their state of activation.

Full evaluation of the importance of sELAM dosage as a prognostic marker of thromboembolic phenomena can only be obtained through a long-term study, with careful follow-up of the patients, which could determine if an increase in sELAM values on diagnosis or (more probably) during disease progress, makes it possible to identify patients with incipient thrombotic risk.

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